

This listing of claims will replace all prior versions and listings of claims in the application:

Listing of Claims:

1. (Currently amended) A method of obtaining avian embryonic germ (EG) cells comprising:

- (i) isolating primordial germ cells (PGCs) from an avian embryo; and
- (ii) culturing said PGCs for a period of at least fourteen days in the absence of a feeder layer in a culture medium comprising

- (1) leukemia inhibitory factor (LIF);
- (2) basic fibroblast growth factor (bFGF);
- (3) stem cell factor (SCF) and
- (4) insulin-like growth factor (IGF),

so that a population of cells comprising avian EG cells is obtained.

2. (Original) The method of Claim 1, wherein the minimal amounts of said growth factors are:

- (1) LIF (0.00625 U/ μ l,
- (2) bFGF (0.25 pg/ μ l),
- (3) IGF (0.5625 pg/ μ l), and
- (4) SCF (4.0 pg/ μ l).

3. (Original) The method of Claim 2, wherein the maximal amounts of said growth factors range from about two times to one hundred times said minimum amounts.

4. (Previously amended) The method of Claim 1, wherein said avian PGCs are obtained from an avian of the order *Gallinaceae*.

5. (Currently amended) The method of Claim 4, wherein said PGCs are chicken PGCs ~~or turkey PGCs~~.

6. (Original) The method of Claim 1, wherein said PGCs are maintained in culture for at least 25 days.

7. (Original) The method according to Claim 6, wherein said PGCs are maintained in culture for longer than 25 days.

8. (Original) The method according to Claim 7, wherein said PGCs are maintained in culture for at least 4 months.

9. (Previously amended) The method of Claim 1, wherein the avian EG cells produce mouse-stage specific antigen 1, and/or react with EMA-1 or MC-480 monoclonal antibody.

10. (Currently amended) The method of Claim 9, wherein ~~the EG phenotype of said cells is further confirmed by transferral of such~~ said avian EG cells are transferred to a suitable avian embryo.

11. (Original) The method of Claim 10, wherein said embryo is a stage X chicken embryo.

12. (Previously amended) The method of Claim 1, which further comprises:
(iii) transfecting or transforming the resultant EG cells with a nucleic acid sequence.

13. (Previously amended) The method of Claim 12, wherein said nucleic acid comprises a nucleotide sequence that encodes a polypeptide and is functionally linked to gene expression regulatory sequences that are operable in an avian cell.

14. (Currently amended) A method of producing chimeric avians comprising:
- (i) isolating primordial germ cells (PGCs) from an avian;
 - (ii) culturing the PGCs for a period of at least fourteen days in the absence of a feeder layer in a tissue culture medium containing at least the following growth factors:
 - (1) leukemia inhibitory factor (LIF);
 - (2) basic fibroblast growth factor (bFGF);
 - (3) stem cell factor (SCF) and
 - (4) insulin-like growth factor (IGF),for a sufficient time to produce embryonic germ (EG) cells;
 - (iii) obtaining a germline and somatic cell chimeric avian.
15. (Previously amended) The method according to Claim 14, wherein said PGCs are derived from avian embryos of the order *Gallinaceae*.
16. (Currently amended) The method according to Claim 15, wherein said avian embryos are ~~turkey~~ or chicken embryos.
17. (Original) The method according to Claim 14, wherein said EG cells are transfected or transformed with a desired nucleic acid sequence prior to transferal to a recipient avian embryo.
18. (Previously amended) The method of Claim 17, wherein said nucleic acid comprises a nucleotide sequence that encodes a polypeptide and is functionally linked to gene expression regulatory sequences that are operable in an avian cell.
19. (Previously amended) The method of Claim 17, wherein said nucleic acid encodes a polypeptide that can be recovered from the systemic circulatory system, body fluids, or tissues of an avian having said nucleic acid sequence.
20. (Original) The method according to Claim 14, wherein the PGCs are injected into the dorsal aorta of a recipient avian embryo or into recipient blastoderms.

21. (Previously amended) The method of Claim 17, wherein said nucleic acid encodes a polypeptide that is a growth factor or an enzyme.

22. (Previously amended) The method of Claim 12, wherein said nucleic acid encodes a polypeptide that can be recovered from the systemic circulatory system, body fluids, or tissues of an avian having said nucleic acid sequence.

23. (Previously amended) The method of Claim 12, wherein said nucleic acid encodes a polypeptide that is a growth factor or an enzyme.

24. (Cancelled).

25. (Previously amended) A method of producing germline chimeric avians comprising:

- (i) isolating primordial germ cells (PGCs) from a Stage XII-XIV avian embryo;
- (ii) maintaining said PGCs for a period of at least fourteen days in a tissue culture medium containing at least the following growth factors:
 - (1) leukemia inhibitory factor (LIF);
 - (2) basic fibroblast growth factor (bFGF);
 - (3) stem cell factor (SCF) and
 - (4) insulin-like growth factor (IGF),
- (iii) transferring PGCs produced by step (ii) into a recipient avian embryo; and
- (iv) obtaining germline chimeric avians.

26. (Previously amended) A method of producing germline and somatic cell chimeric avians which comprises:

- (i) isolating primordial germ cells (PGCs) from a Stage XII-XIV avian embryo;
- (ii) maintaining said PGCs in a tissue culture medium containing at least the

following growth factors:

- (1) leukemia inhibitory factor (LIF);
- (2) basic fibroblast growth factor (bFGF);
- (3) stem cell factor (SCF) and
- (4) insulin-like growth factor (IGF),

for a sufficient time to produce embryonic (EG) cells;

(iii) transferring cells produced by step (ii) comprising said EG cells into recipient avian embryo of the same species as the avian used to obtain said isolated PGCs;

(iv) allowing said recipient avian embryo containing said transferred EG cells to develop into a germline and somatic cell chimeric avian.

27. (Previously amended) A method for producing avian embryonic (EG) cells comprising:

- (i) isolating primordial germ cells (PGCs) from a Stage XII-XIV avian embryo;
- (ii) culturing said PGCs for a period of at least fourteen days in tissue culture in the

absence of a feeder layer in a culture medium comprising:

- (1) leukemia inhibitory factor (LIF);
- (2) basic fibroblast growth factor (bFGF);
- (3) stem cell factor (SCF) and
- (4) insulin-like growth factor (IGF),

so that a population of cells comprising avian EG cells is produced.

28. (Previously amended) A method for producing a germline chimeric avian comprising:

- (i) isolating primordial germ cells (PGCs) from a Stage XII-XIV avian embryo;
- (ii) culturing said PGCs for a period of at least fourteen days in tissue culture in the absence of a feeder layer in a culture medium comprising:
 - (1) leukemia inhibitory factor (LIF);
 - (2) basic fibroblast growth factor (bFGF);
 - (3) stem cell factor (SCF) and
 - (4) insulin-like growth factor (IGF);
- (iii) transferring said PGCs produced by step (ii) into a recipient avian embryo of the same species as the avian used to obtain said isolated PGCs;
- (iv) allowing said recipient avian embryo containing said transferred PGCs to develop into a germline chimeric avian.

29. (Previously amended) A method for producing a germline chimeric avian comprising:

- (i) isolating primordial germ cells (PGCs) from a Stage XII-XIV avian embryo;
- (ii) culturing said PGCs for a period of at least fourteen days in tissue culture in the absence of a feeder layer in a culture medium comprising:
 - (1) leukemia inhibitory factor (LIF);
 - (2) basic fibroblast growth factor (bFGF);
 - (3) stem cell factor (SCF) and
 - (4) insulin-like growth factor (IGF);
- (iii) transferring said PGCs produced by step (ii) into a recipient avian embryo of the same species as the avian used to obtain said isolated PGCs; and
- (iv) allowing said recipient avian embryo containing said transferred PGCs to develop into a germline chimeric avian.

30. (Previously amended) A method for producing germline or somatic cell chimeric avians comprising:

(i) isolating primordial germ cells (PGCs) from a Stage XII-XIV avian embryo;
(ii) culturing said PGCs for a period of at least fourteen days in tissue culture in the absence of a feeder layer in a culture medium comprising:

- (1) leukemia inhibitory factor (LIF);
- (2) basic fibroblast growth factor (bFGF);
- (3) stem cell factor (SCF) and
- (4) insulin-like growth factor (IGF);

for a sufficient time to produce embryonic germ (EG) cells;

(iii) transferring said cells produced by step (ii) comprising EG cells into a recipient avian embryo of the same species as the avian used to obtain said isolated PGCs; and

(iv) allowing said recipient avian embryo containing said transferred EG cells to develop into a germline or somatic cell chimeric avian.